PROCEEDINGS OF THE SOCIETY FOR EXPERIMENTAL BIOLOGY AND MEDICINE 173, 553–563 (1983)

# Glucose and Lactate Oxidation Rates in the Fetal Lamb (41686)

WILLIAM W. HAY, JR.,[1] STEPHEN A. MYERS, JOHN W. SPARKS, RANDALL B. WILKENING, GIACOMO MESCHIA, AND FREDERICK C. BATTAGLIA

*Division of Perinatal Medicine and the Departments of Pediatrics, Physiology, and Obstetrics and Gynecology, University of Colorado School of Medicine, 4200 East Ninth Avenue, Denver, Colorado 80262*

*Abstract.* Both glucose and lactate are nutrients of the ovine fetus. Each may be used by the fetus as a fuel for oxidation or as a source of carbon for energy storage and net tissue accretion. The present report describes the oxidation rates of glucose and lactate *in vivo* for the fetal lamb over a relatively short time period. The fraction of fetal glucose or lactate oxidized was defined as the ratio of $^{14}CO_2$ excretion across the umbilical circulation to the net entry of [$^{14}$C]glucose or [$^{14}$C]lactate into fetal tissues. The fraction of glucose oxidized over a 3-hr study averaged 61.2%, accounting for 2.55 $mg \cdot min^{-1} \cdot kg^{-1}$ of glucose oxidized and for 28% of the simultaneous net oxygen uptake. The fraction of lactate oxidized averaged 71.5%, accounting for 4.12 $mg \cdot min^{-1} \cdot kg^{-1}$ of lactate oxidized. Oxidation fractions and rates for both glucose and lactate increased with their concentrations in fetal blood suggesting sparing of other fuels for oxidation at higher glucose and lactate concentrations.

Several studies have demonstrated net umbilical uptakes of glucose and lactate by the unstressed fetal lamb during the last month of gestation. These substrates account for approximately 40% of total calorie and 45% of total carbon uptakes (1–3). It has been assumed that glucose is used by the fetus primarily for oxidation. However, tracer studies of both glucose and lactate oxidation in adult animals and human have demonstrated oxidation fractions significantly less than 1.00 (4–6). In a growing animal the rates of glucose and lactate oxidation might be less than in the adult because these substrates could provide carbon for net tissue accretion. In order to quantify the partition of fetal glucose and lactate for oxidation and for other metabolic fates, we have measured glucose and lactate oxidation rates in fetal lambs under *in vivo* steady state conditions.

**Materials and Methods.** *1. Surgical preparation and experimental protocol.* Columbia–Rambouillet, mixed-breed ewes were studied between 120 and 140 days gestation (term = 147 days). Surgery was performed with pentobarbital sedation and pontocaine spinal anesthesia (6 mg in 10% dextrose). Polyvinyl catheters were placed in a fetal pedal artery (advanced to the midabdominal aorta), a fetal pedal vein (advanced to the femoral vein), and a fetal umbilical vein (positioned for sampling in the common umbilical vein, distal to the portal sinus). The ewes recovered for about 1 week before study and were fed a standard diet of hay, alfalfa pellets mixed with Omolene (grain mix, Farmer's Market Assoc., Denver), and water *ad libitum.*

The study protocol consisted of 150- to 180-min primed-constant infusions of [$^{14}$C]glucose (labeled universally or at the first or sixth carbon, Amersham) or [U-$^{14}$C]lactate (New England Nuclear). The tracers were infused into a fetal femoral vein. This site of injection provides a fairly uniform specific activity in fetal arterial blood (1) but cannot avoid differences in intracellular and venous specific activities, especially for lactate (3). These differences make the calculation of absolute lactate oxidation rates dependent on the site of sampling (9) with arterial sampling providing a minimum rate, but have no effect on the calculation of fractional oxidation rates. Umbilical blood flow was measured by the antipyrine steady-state diffusion technique (7, 8). After a 60-min equilibration period, whole blood samples were taken simultaneously from the

[1] To whom reprint requests should be addressed at the Department of Pediatrics.



0037-9727/83 $1.50
Copyright © 1983 by the Society for Experimental Biology and Medicine.
All rights reserved.

fetal artery and umbilical vein at 60, 90, 120, 150, and occasionally 180 min. The blood samples were analyzed for glucose and lactate concentrations, [$^{14}$C]glucose or [$^{14}$C]lactate concentrations, $^{14}CO_2$ concentration, oxygen content, and antipyrine concentration.

*2. Analytical methods.* Whole blood was collected in plastic syringes and an aliquot was deproteinized immediately with zinc sulfate and barium hydroxide for glucose analysis by glucose oxidase (Sigma). [$^{14}$C]glucose concentration was measured as previously described (1). Two identical whole blood protein-free filtrate aliquots were incubated at 37°C for 1 hr, one with water and one with glucose oxidase (converting glucose to gluconic acid). Each aliquot was passed through an anion exchange column (200–400 mesh, formate form). Glucose was retained on one column as gluconic acid but not on the other column. The difference in radioactive counts between the column eluates represents the counts specific to glucose.

Another aliquot of whole blood was collected in frozen syringes, deproteinized immediately in perchloric acid, and assayed for lactate concentration by a fluorometric assay using lactate dehydrogenase (10). Radioactive lactate concentration and specific activity were measured as previously described using a fluoride form anion exchange column (3). Lactate was eluted with 2 *N* acetic acid after a 0.3 *N* acetic acid wash. Aliquots of the eluate were counted for radioactivity and analyzed for lactate concentration. Oxygen content was measured from NaF coated capillaries by a Lex-$O_2$-Con instrument (Lexington Instruments, Waltham, Mass.). Antipyrine was measured in whole blood with a Technicon Auto Analyzer (7, 8).

Blood for $^{14}CO_2$ analysis was collected anaerobically in iced, oiled, glass syringes. The needle tips were sealed with rubber stoppers. The blood was injected anaerobically (through a plastic cap, sealed with modeling clay or through a tight-fitting rubber stopper) into a plastic cup suspended inside a standard liquid scintillation vial. The vials were weighed before and after addition of the blood to determine the quantity of blood in each vial. An approximately equal volume of 1 *N* HCl was then added to the blood anaerobically and the vials incubated for 5–24 hr. Initially, the acid-released $CO_2$ was trapped in the bottom of the vial in 0.3 *N* $Ba(OH)_2$. However, in all later experiments Protosol, a 0.5 *M* quaternary ammonium hydroxide solution (New England Nuclear), was used to trap the $CO_2$. After incubation, the cup containing blood and acid was removed. Scintillation cocktail (PCS, Amersham) was added directly to the vials containing $Ba(OH)_2$; Econofluor (New England Nuclear) with 0.5% methanol was added to the vials containing Protosol and the vials counted. Control experiments using $NaH^{14}CO_3$ with a known radioactivity were used to determine overall recovery. Protosol increased the recovery from 25% for $Ba(OH)_2$ to about 90–95%, trapping the $CO_2$ as a stable carbamate in the relatively acidic scintillation cocktail.

*3. Model and calculations.* The model employed in this study for measuring glucose or lactate oxidation rates is shown in Fig. 1. The Equations are presented in Table I. During a tracer infusion into the fetal circulation, there is a net loss of tracer down its concentration gradient, into the placenta and mother. For substrates with a high placental permeability, such as glucose, a large fraction (average = 50% for glucose) of the tracer infused into the fetus is lost in this way without entering fetal tissues for metabolism (1). A larger fraction of other substrates, such as lactate, to which the placenta is less permeable, remain within the fetus. Only about 10% of tracer lactate infused into the fetus is lost to the placenta and about 90% remains for metabolism by fetal tissues (3).

The remainder of the tracer which enters the fetus (net fetal tracer uptake rate) can be used by fetal tissues for metabolism. We define the oxidation fraction as the ratio of the net $^{14}CO_2$ flux to the uteroplacenta from the fetus to the net fetal tracer uptake rate (Eq. 1).

The net tracer glucose or tracer lactate flux to the uteroplacenta from the fetus and the net $^{14}CO_2$ flux to the uteroplacenta from the fetus were calculated as the product of umbilical blood flow and the umbilical arterial-venous concentration differences for tracer glucose, tracer lactate, and $^{14}CO_2$, respectively (Eq. 2, 3). The rate of substrate utilization was calculated as the net fetal tracer uptake rate

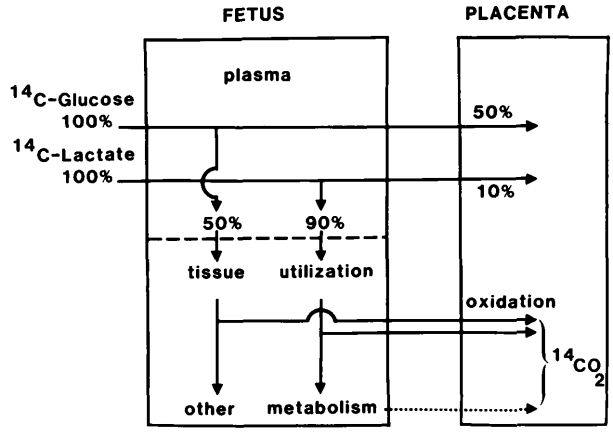


FIG. 1. Model for measuring fetal substrate oxidation rates. Labeled fetal $CO_2$ production is measured as the umbilical $^{14}CO_2$ excretion rate; to calculate the fraction of substrate oxidized, labeled $CO_2$ excretion is compared with net fetal tissue utilization of tracer.

divided by the steady-state fetal arterial substrate specific activity (Eq. 4). The amount of substrate oxidized was then calculated as the product of the fraction of substrate oxidized and the rate of substrate utilized (Eq. 5).

Steady-state was defined as a concentration of tracer ([$^{14}$C]glucose or [$^{14}$C]lactate) and tracee (glucose or lactate) which had a variability of less than 5% during the constant tracer infusion.

Net fetal glucose, lactate, and oxygen uptakes were calculated as the product of umbilical blood flow and umbilical venous–arterial concentration difference for each sub-

TABLE I

| Equation No. | |
|---|---|
| 1 | Glucose or lactate oxidation fraction $= \frac{\text{Net } ^{14}CO_2 \text{ flux to uteroplacenta, from fetus (dpm/min)}}{\text{Net fetal tracer [}^{14}\text{C]glucose or [}^{14}\text{C]lactate uptake (dpm/min)}}$ |
| 2 | Net $^{14}CO_2$ flux to uteroplacenta, from fetus (dpm/min) = Umbilical blood flow (ml/min) × umbilical arteriovenous $^{14}CO_2$ difference (dpm/ml) |
| 3 | Net fetal tracer [$^{14}$C]glucose or [$^{14}$C]lactate uptake (dpm/min) = [Tracer infusion rate (dpm/min)] − [umbilical blood flow (ml/min) × umbilical arteriovenous tracer difference (dpm/ml)] |
| 4 | Fetal substrate utilization rate (mg/min) = Net fetal tracer uptake (dpm/min) ÷ fetal arterial substrate specific activity (dpm/mg) |
| 5 | Glucose or lactate oxidation rate (mg/min) = Oxidation fraction × glucose or lactate utilization rate (mg/min) |
| 6 | Fraction of oxygen uptake for glucose oxidation $= \frac{6 \times \text{glucose oxidized (m}M\text{/min)}}{\text{Net oxyen uptake (m}M\text{/min)}}$ |
| 7 | Fraction of oxygen uptake for lactate oxidation $= \frac{3 \times \text{Lactate oxidized (m}M\text{/min)}}{\text{Net oxygen uptake (m}M\text{/min)}}$ |
| 8 | Amount of oxygen uptake for glucose or lactate oxidation (m$M$/min) = fraction of oxygen for glucose or lactate oxidation × net oxygen uptake (m$M$/min) |

strate. The fraction of fetal oxygen uptake used for glucose oxidation was calculated as 6 times the molar ratio of glucose oxidized to the net oxygen uptake (Eq. 6), and for lactate, as 3 times the molar ratio of lactate oxidized to the net oxygen uptake (Eq. 7). The amount of oxygen taken up by the fetus and used for glucose or lactate oxidation was calculated as the product of the net oxygen uptake and the fraction of oxygen used for oxidation of either glucose or lactate (Eq. 8).

Plasma glucose concentrations were estimated from whole blood glucose according to the equation:

$$P = \frac{W + 3.3H}{1 - 0.24H}$$

where $P$ = plasma glucose (mg/dl), $W$ = whole blood glucose (mg/dl), and $H$ = hematocrit expressed as a fraction. This equation was developed empirically from previous measure-

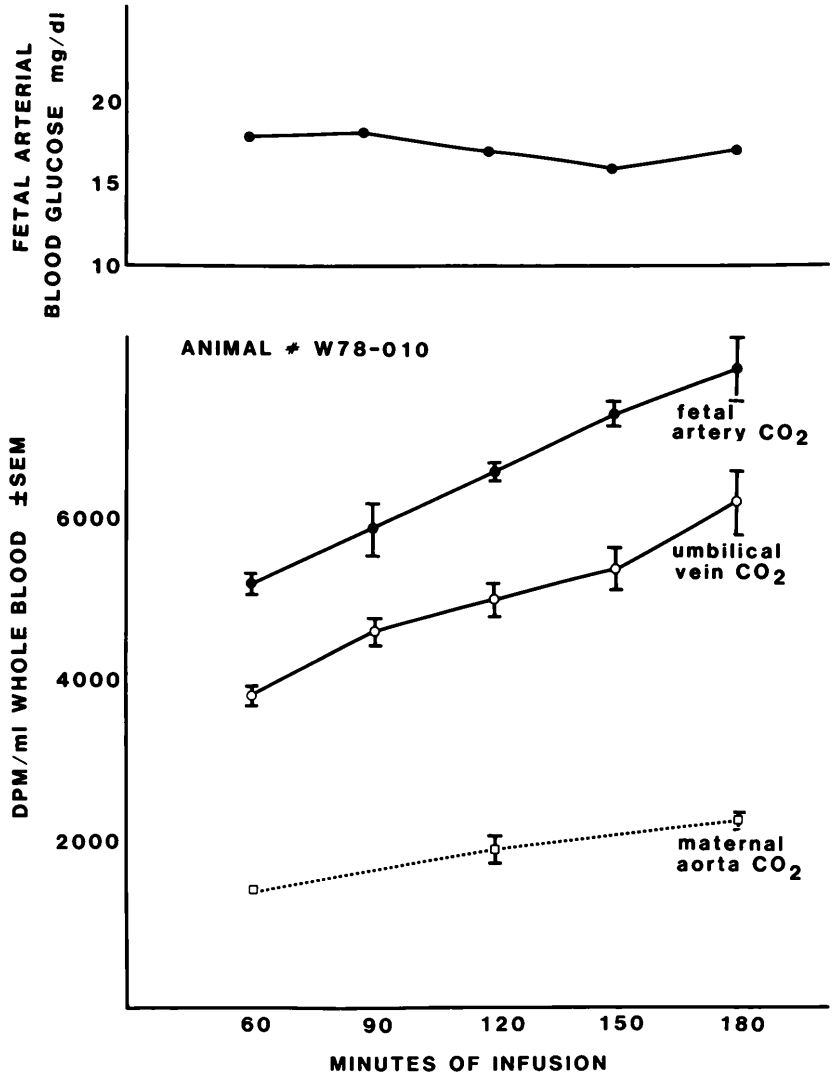


FIG. 2. Blood glucose concentrations (mg/dl) and blood $^{14}CO_2$ concentrations (dpm/ml) in the umbilical artery and vein and the maternal aorta in one animal.

ments in sheep of fetal blood glucose, plasma glucose, and hematocrit in our laboratory.

**Results.** Umbilical arteriovenous $^{14}CO_2$ concentration differences did not change significantly after the first hour of the infusion ($P > 0.1$), despite increasing $^{14}CO_2$ concentration in fetal and maternal blood. Figure 2 presents the $^{14}CO_2$ concentration in fetal and maternal vessels for a single animal. Since $^{14}CO_2$ was accumulating slowly in the fetus, comparison of umbilical excretion of $^{14}CO_2$ with net fetal [$^{14}$C]glucose or [$^{14}$C]lactate uptake provides a minimum rate of oxidation. Additional $^{14}CO_2$ produced from [$^{14}$C]-glucose or [$^{14}$C]lactate is contained in the enlarging fetal $^{14}CO_2$ pool. An estimation of the rate of $^{14}CO_2$ accumulation (slope of $^{14}CO_2$ concentration in disintigrations per minute per milliliter per minute × weight of the fetus × estimated $HCO_3$ space in milliliters per kilogram of fetal body weight) would increase the $^{14}CO_2$ production rate (and thus the oxidation rate) by about 6%. Because the $HCO_3$ space in these animals was estimated from chloride dilutions and not directly measured, the 6% correction has not been applied to the present data.

Table II presents data for net glucose fluxes as well as [$^{14}$C]glucose and $^{14}CO_2$ fluxes for the studies of fetal glucose oxidation. In the 13 experiments with [U-$^{14}$C]glucose the fraction of glucose oxidized averaged 61.2 ± 4.0% (SEM) of net fetal glucose utilization, accounting for 2.55 $mg \cdot min^{-1} \cdot kg^{-1}$. This amount of glucose oxidized accounted for 28% (0.089 $mmole \cdot min^{-1} \cdot kg^{-1}$) of the simultaneously measured fetal oxygen uptake.

A wide range of fetal arterial plasma glucose concentrations was observed, occurring spontaneously or in response to fasting of the ewe. The glucose oxidation fraction (Fig. 3) increased with fetal plasma glucose concentration ($R = 0.602$, $P < 0.05$). Thus, the fetal glucose oxidation rate (Fig. 4) increased directly with fetal glucose concentration ($R = 0.726$, $P < .01$). The fraction of net oxygen uptake used for glucose oxidation was also directly related to glucose concentration ($R = 0.618$, $P < 0.05$), due to the fact that there was not a significant relationship between oxygen uptake and glucose concentration. In the six experiments with [1-$^{14}$C]glucose the mean glucose oxidation fraction was 61.8% (±3.6% SEM) at a mean plasma glucose of 20.0 ± 1.5 mg/dl. In the three experiments with [6-$^{14}$C]glucose the mean glucose oxidation fraction was 40% (range 29 to 51) at a mean plasma glucose of 12.5 mg/dl (range 11.5 to 13.3).

Table III presents data for the six fetal lactate oxidation studies. The lactate oxidation fraction averaged 0.715 ± 0.07 accounting for 4.12 $mg \cdot min^{-1} \cdot kg^{-1}$ of lactate utilization.

Lactate utilization tended to increase directly with fetal arterial plasma lactate concentration, as did the fraction of lactate oxidized (Fig. 5, $R = 0.77$, $P < 0.1$). As with glucose, oxygen uptake did not vary significantly with lactate concentration. The lactate oxidation rate varied directly with lactate concentration (Fig. 6, $R = 0.89$, $P < 0.05$).

The amount of oxygen required for lactate oxidation (0.137 $mmole \cdot min^{-1} \cdot kg^{-1}$) includes oxygen used for oxidation of lactate derived from net fetal lactate uptake in the umbilical circulation, from fetal glucose, and from nonglucose, lactate precursors. In a previous report, we estimated that approximately half of the lactate used by the fetus was derived from fetal glucose, and the other half from nonglucose, lactate precursors, and net umbilical lactate uptake (3). Therefore, the combined oxidation of fetal glucose and fetal lactate derived from nonglucose precursors could account for approximately 0.16 $mmole \cdot min^{-1} \cdot kg^{-1}$ of oxygen which would represent about 50% of fetal oxygen consumption.

**Discussion.** Previous reports from our laboratory have described the umbilical glucose:oxgyen quotient as one index of fetal glucose metabolism (11). The umbilical glucose:oxygen quotient represents the fraction of net fetal oxygen uptake that would be used for oxidation of net glucose uptake via the umbilical circulation, assuming that all of this glucose is oxidized. In the fed-state, with normal glucose concentrations, the umbilical glucose:oxygen quotient is about 0.5, decreasing with fasting-induced hypoglycemia to about 0.2. Fed-state umbilical lactate:oxygen quotients have averaged about 0.21, decreasing to about 0.13 during fasting-induced hypoglycemia, coincident with a decrease in fetal plasma lactate concentration from about 1.7

TABLE II. OXIDATION OF GLUCOSE

| Animal No. | Fetal arterial plasma glucose (mg/dl) | Net fetal glucose uptake ($mg \cdot min^{-1} \cdot kg^{-1}$) | Fetal glucose utilization ($mg \cdot min^{-1} \cdot kg^{-1}$) | Fraction of glucose oxidized | Fetal glucose oxidation rate ($mg \cdot min^{-1} \cdot kg^{-1}$) | Net fetal oxygen uptake ($mM \cdot min^{-1} \cdot kg^{-1}$) | Oxygen for oxidation of glucose utilization ($mM \cdot min^{-1} \cdot kg^{-1}$) | Fraction of oxygen for oxidation of glucose utilization | Umbilical glucose: oxygen quotient |
|---|---|---|---|---|---|---|---|---|---|
| 058-1-c | 15.15 | 3.55 | 6.19 | 0.63 | 3.40 | 0.313 | 0.130 | 0.42 | 0.38 |
| 076-2-c | 24.06 | 4.82 | 3.34 | 0.48 | 1.60 | 0.400 | 0.053 | 0.13 | 0.41 |
| 010-1-c | 19.54 | 5.05 | 5.23 | 0.68 | 3.56 | 0.345 | 0.119 | 0.34 | 0.49 |
| 018-1-c | 19.01 | 3.31 | 4.88 | 0.64 | 3.12 | 0.275 | 0.104 | 0.38 | 0.40 |
| 023-1-d | 13.32 | 1.53 | 4.42 | 0.51 | 2.25 | 0.213 | 0.075 | 0.35 | 0.24 |
| 038-1-d | 11.54 | 2.35 | 5.74 | 0.40 | 2.30 | 0.265 | 0.077 | 0.29 | 0.30 |
| 064-1-c | 22.40 | 5.37 | 7.60 | 0.66 | 5.02 | 0.374 | 0.167 | 0.45 | 0.48 |
| 074-1-e | 27.39 | 4.35 | 4.67 | 0.65 | 3.04 | 0.335 | 0.101 | 0.30 | 0.43 |
| 086-1-e | 16.74 | 2.67 | 7.27 | 0.40 | 2.91 | 0.312 | 0.097 | 0.31 | 0.26 |
| 079-1-e | 10.13 | 2.74 | 2.90 | 0.62 | 1.80 | 0.331 | 0.060 | 0.18 | 0.27 |
| 080-1-e | 11.62 | 4.01 | 3.19 | 0.63 | 2.01 | 0.351 | 0.067 | 0.19 | 0.40 |
| 087-2a-e | 14.06 | 3.02 | 3.08 | 0.55 | 1.69 | 0.330 | 0.056 | 0.17 | 0.67 |
| 087-2b-e | 13.00 | 0.82 | 3.21 | 0.54 | 1.73 | 0.374 | 0.058 | 0.15 | 0.07 |
| 019-2-e | 19.92 | 4.02 | 3.95 | 0.48 | 1.90 | 0.280 | 0.063 | 0.23 | 0.48 |
| 036-1a-e | 24.70 | 3.89 | 4.80 | 0.80 | 3.84 | 0.229 | 0.128 | 0.56 | 0.57 |
| 036-1b-e | 11.59 | 0.17 | 3.21 | 0.57 | 1.83 | 0.229 | 0.061 | 0.27 | 0.02 |
| 061-1-e | 13.58 | 1.92 | 7.46 | 0.43 | 3.21 | 0.291 | 0.107 | 0.37 | 0.19 |
| 062-1a-e | 21.16 | 4.37 | 5.04 | 0.69 | 3.48 | 0.371 | 0.116 | 0.31 | 0.42 |
| 062-1b-e | 13.88 | 2.48 | 3.59 | 0.66 | 2.37 | 0.360 | 0.079 | 0.22 | 0.22 |
| 052-1-d | 12.61 | 0.42 | 4.19 | 0.29 | 1.22 | 0.328 | 0.041 | 0.12 | 0.04 |
| 080-1-e | 29.00 | 4.42 | 3.67 | 0.93 | 3.41 | 0.321 | 0.114 | 0.35 | 0.46 |
| $N = 22$ | | | | | | | | | |
| $\bar{X}$ | 17.36 | 3.10 | 4.65 | 0.583 | 2.65 | 0.316 | 0.089 | 0.29 | 0.34 |
| SEM | 1.44 | 0.33 | 0.32 | 0.032 | 0.21 | 0.011 | 0.007 | 0.02 | 0.04 |

*Note:* 1 = singleton pregnancy, 2 = twin pregnancy, a = fed study, b = fasted study, c = $^{14}C$ label on C-1, d = $^{14}C$ label on C-6, e = $^{14}C$ label on all 6 carbons.

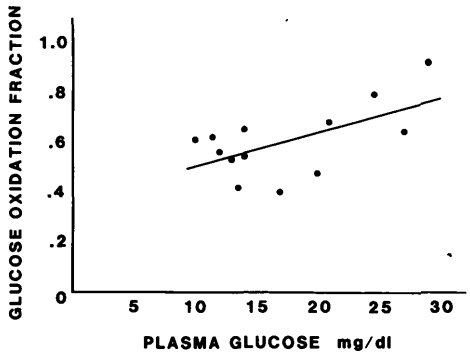


FIG. 3. Glucose oxidation fractions for the 13 animals infused with [U-$^{14}$C]glucose: $R = 0.602$, $P < 0.05$.

to about 1.2 m$M$ (3). From these studies, it was concluded that if completely oxidized, glucose and lactate could account for a large fraction of ovine, fetal oxidative metabolism in the fed-state. The present study provides the first quantitative demonstration to support the hypothesis that glucose and lactate are important substrates of fetal oxidative metabolism, by showing that the oxidation rate of these two substrates over a short time period (1–3 hr) can account for about 50% of fetal $O_2$ consumption.

Previous studies have concluded that immediate and "long term" oxidation rates of glucose are not well separated in time (12). Such a conclusion was based on the observation in rats that the specific activity of expired $CO_2$ relative to plasma glucose specific activity continues to increase markedly in the first 4 to 6 hr of a tracer glucose infusion before reaching a plateau. In the present experiments, a progressive contribution of "long term" fates of glucose metabolism to oxidation should have increased the $^{14}CO_2$ umbilical arterial–venous difference. However, there was no discernable increase in the umbilical arterial–venous $CO_2$ difference in the third hour of the infusion compared to the second hour (paired $t$ test, $P = 0.8$), suggesting that "long term" glucose and lactate oxidation did not appreciably affect the measurement of "short term" oxidation rates.

Several reports in the literature describe glucose and lactate oxidation fractions for rats (13–17), rabbits (18), dogs (4, 5, 19–21), goats (22), sheep (23–25), and humans (6, 26). Re-

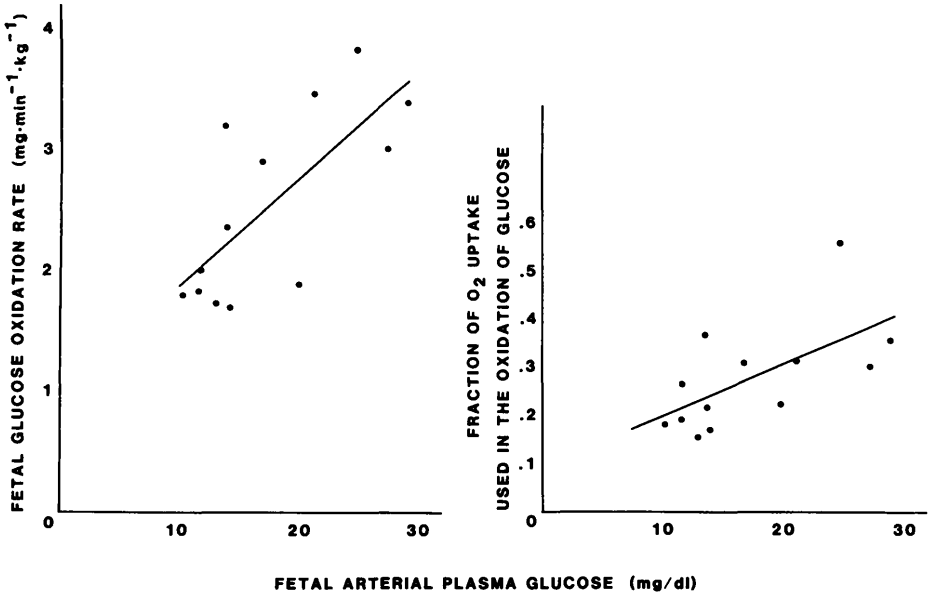


FIG. 4. Left: fetal glucose oxidation rate for the 13 animals infused with [U-$^{14}$C]glucose are compared with the fetal arterial plasma glucose concentration, $R = 0.726$, $P < 0.01$. Right: fraction of net oxygen uptake used for the oxidation of the glucose compared with the fetal arterial plasma glucose concentration, $R = 0.618$, $P < 0.05$.

TABLE III. OXIDATION OF LACTATE

| Animal No. | Fetal arterial plasma lactate (mg/dl) | Net fetal lactate uptake ($mg \cdot min^{-1} \cdot kg^{-1}$) | Fetal lactate utilization ($mg \cdot min^{-1} \cdot kg^{-1}$) | Fraction of lactate oxidized | Letal lactate oxidation ($mg \cdot min^{-1} \cdot kg^{-1}$) | Net fetal oxygen uptake ($mM \cdot min^{-1} \cdot kg^{-1}$) | Oxygen for lactate oxidation ($mM \cdot min^{-1} \cdot kg^{-1}$) | Fraction of oxygen for lactate oxidation | Umbilical lactate/$O_2$ quotient |
|---|---|---|---|---|---|---|---|---|---|
| 066-1 | 10.97 | 1.46 | 8.34 | 0.45 | 3.75 | 0.288 | 0.125 | 0.43 | 0.17 |
| 021-1 | 13.00 | 2.11 | 4.57 | 0.87 | 3.98 | 0.324 | 0.133 | 0.31 | 0.22 |
| 030-1 | 12.92 | 0.17 | 5.32 | 0.61 | 3.24 | 0.238 | 0.108 | 0.45 | 0.17 |
| 042-1 | 14.91 | 0.18 | 5.67 | 0.91 | 5.17 | 0.259 | 0.172 | 0.67 | 0.18 |
| 063-1 | 15.89 | 1.57 | 7.42 | 0.81 | 5.98 | 0.304 | 0.199 | 0.66 | 0.13 |
| 070-1 | 11.29 | 0.11 | 4.03 | 0.64 | 2.58 | 0.242 | 0.086 | 0.36 | 0.11 |
| $\bar{X}$ | 13.16 | 1.44 | 5.89 | 0.72 | 4.12 | 0.276 | 0.137 | 0.50 | 0.16 |
| SEM | 0.79 | 0.17 | 0.68 | 0.07 | 0.51 | 0.014 | 0.017 | 0.05 | 0.02 |

ported glucose oxidation fractions range from 0.31 to 0.75; reported lactate oxidation fractions range from 0.36 to 0.90. These values are not strictly comparable to the present data because of two differences: one, in physiologic states (e.g., handling stress, anesthesia, fasting of different degrees) and another in methodologic differences for measuring oxidation rates (e.g., steady-state $^{14}CO_2$ to $^{14}C$-labeled substrate specific activity ratios, ratio of $^{14}CO_2$ appearance in expired air to the rate of $^{14}C$-labeled substrate infusion). Nevertheless, several of these reports show a decrease in glucose oxidation with fasting, which is consistent with the present study. Independent effects of both insulin and glucose on enhancing glucose oxidation have been demonstrated *in vitro* in adipocytes (27, 28). In diabetic animals glucose oxidation is depressed but can return to normal levels with insulin therapy (6). Glucose and insulin concentrations decrease during fasting in both maternal and fetal sheep (29). Bergman has presented data from both pregnant and nonpregnant ewes, studied under chronic, conscious conditions (24, 25). Glucose oxidation fraction averaged 0.31 in nonpregnant ewes and was unchanged by fasting. However, in twin-pregnant sheep, plasma glucose concentration decreased by 38% (from 47 ± 3 to 29 ± 3 mg/dl), associated with a 16% decrease in glucose oxidation fraction from 0.37 to 0.31.

The present studies confirm previous investigations (30) demonstrating a normal rate of fetal oxidative metabolism during fasting. Presumably, the rate of oxidative metabolism is relatively fixed, whereas individual substrate oxidation rates are dependent on the specific substrate concentration and the coincident concentration of other substrates and the hormonal milieu.

At relatively high glucose and lactate concentrations, the fraction of oxidation of each substrate increases. As oxygen uptake does not increase with glucose and lactate concentrations, glucose and lactate can spare the oxidation of other substrates. In the fetal lamb, fat uptake (31) and accretion are not large (fat content at term is about 3% of body weight compared to 18% for humans (32)). Thus, the "sparing" effect of glucose and lactate may apply primarily to amino acids.

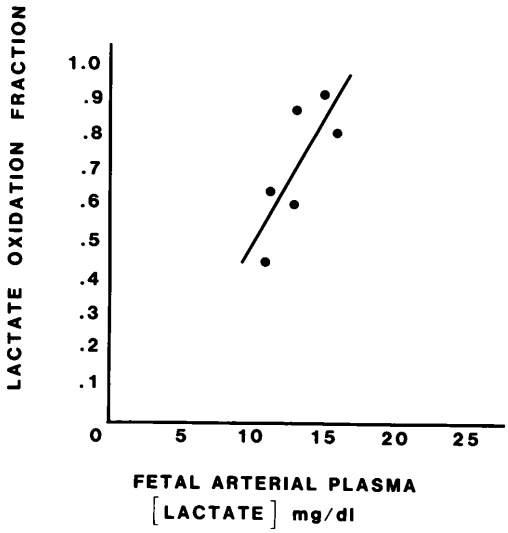


FIG. 5. Lactate oxidation fractions for the six animals infused with [$^{14}$C]lactate: $R = 0.77$, $P < 0.01$.

At low glucose and lactate concentrations, relatively large fractions of glucose and lactate are not oxidized. Because oxygen uptake is not changed, other substrates would be preferentially oxidized.

The fetal oxygen uptake not used for the short term oxidation of glucose and lactate must be accounted for by the oxidation of other substrates. Glucose itself, through slow turnover of intermediate products (e.g., fructose) may contribute carbon for this additional rate of oxidation. Although not taken up in large amounts by the umbilical circulation of the fetal lamb, fatty acids, ketoacids, and acetate may contribute carbon for oxidation in the fetus (31, 33, 34). However, we think it more likely that amino acids represent the bulk of the substrates that serve as alternate fuels for the fetus.

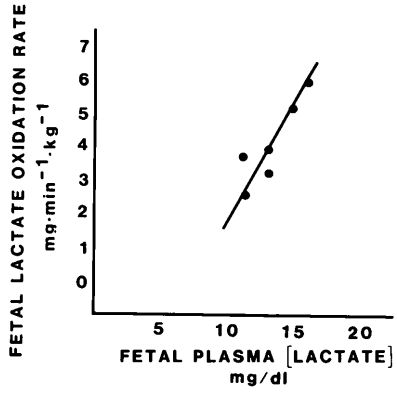


FIG. 6. Fetal lactate oxidation rate versus fetal arterial plasma lactate concentration: $R = 0.89$, $P < 0.05$.

In the fed state, amino acid uptake exceeds the requirements for tissue accretion by approximately 50%, ranging from 20% for the basic amino acids, histidine and lysine, to 65–75% for neutral amino acids such as alanine, serine, threonine, and glutamine/glutamate (35, 36). The measurement of ovine fetal amino acid uptake made in this laboratory has accounted for approximately 3.94 g of carbon · day$^{-1}$ · kg$^{-1}$ (36). Complete oxidation of the fraction (about 50%) not used for accretion (at 2 mole of oxygen consumed per mole of carbon) could account for approximately 0.2 mmole $O_2 \cdot min^{-1} \cdot kg^{-1}$, or two times the difference between the oxygen uptake and the oxygen used for oxidation of glucose and lactate in the present studies. Thus, there is sufficient excess of amino acid uptake by the fetus to satisfy that portion of oxygen uptake which is not used for "short term" oxidation of glucose and lactate. The actual contribution of amino acids to oxidation has only been measured for lysine (37). In normoglycemic fetal lambs, the lysine oxidation fraction was approximately 9%.

In conclusion, the present report offers direct evidence to support the hypothesis that glucose and lactate are utilized primarily for oxidation in the late gestation ovine fetus, and that the oxidation of these two substrates depends on their concentrations. Since oxygen consumption does not vary appreciably with glucose or lactate concentration, we surmise that at higher concentrations, glucose and lactate can spare the oxidation of other substrates, such as amino acids. The converse of this conclusion, namely, that at lower glucose and lactate concentrations, obligatory nonoxidative requirements of glucose and lactate exceed their oxidative fates, bear further consideration and study.

This work was supported in part by NIH Program Grant HD00781 and NIH Project Grant HD01866. Dr. Hay is the recipient of a Special Emphasis Research Career Award (Diabetes Mellitus: Pediatric Aspects), NIH-NIADDK/

NICHD. Dr. Myers was supported by NIH Training Grant HD 07186.


1. Hay WW Jr, Sparks JW, Quissell BJ, Battaglia FC, Meschia G. Simultaneous measurements of umbilical uptake, fetal utilization rate, and fetal turnover rate of glucose. Amer J Physiol **240**:E662–E668, 1981.
2. James EJ, Raye JR, Gresham EL, Makowski EL, Meschia G, Battaglia FC. Fetal $O_2$ consumption, $CO_2$ production, and glucose uptake in a chronic sheep preparation. Pediatrics **50**:361–1371, 1972.
3. Sparks J. W, Hay WW Jr, Bonds D, Meschia G, Battaglia, FC. Simultaneous measurements of lactate turnover rate and umbilical lactate uptake in the fetal lamb. J Clin Invest 70:179–192, 1982.
4. Depocas F, Minnaire Y, Chatonnet J. Rates of formation and oxidation of lactic acid in dogs at rest and during moderate exercise. Canad J Physiol Pharmacol **47**:603–610, 1969.
5. Feller DD, Chaikoff IL, Strisower EH, Searle GL. Glucose utilization in the diabetic dog, studied with $C^{14}$ glucose. J Biol Chem **188**:865–880, 1951.
6. Manougian E, Pollycove M, Linfoot JA, Lawrence JH. $C^{14}$ Glucose kinetic studies in normal, diabetic, and acromegalic subjects. J Nuclear Med **5**:763–795, 1965.
7. Meschia G, Cotter JR, Makowski EL, Barron DH. Simultaneous measurement of uterine and umbilical blood flows and oxygen uptakes. Q J Exp Physiol **52**:1–18, 1967.
8. Wilkening RB, Anderson S, Martensson L, Meschia G. Placental blood flow as a function of uterine blood flow. Amer J Physiol **242**:H429–H436, 1982.
9. Katz J, Okajima F, Chenowith M, Dunn A. The determination of lactate turnover *in vivo* with $^3H$- and $^{14}C$-labelled lactate. Biochem J **194**:513–524, 1981.
10. Olsen C. An enzymatic fluorometric micromethod for the determination of acetoacetate, beta-hydroxybutyrate, pyruvate and lactate. Clin Chem Acta **33**:293–300, 1971.
11. Tsoulos NG, Colwill JR, Battaglia FC, Makowski EL, Meschia G. Comparison of glucose, fructose, and $O_2$ uptakes by fetuses of fed and starved ewes. Amer J Physiol **221**:234–237, 1971.
12. Stetten D Jr. Glucose synthesis and oxidation in the alloxan-diabetic and cortisone treated rat. In: Brookhaven Symposia in Biology, No. 5, Major Metabolic Fuels. Upton, N.Y., Assoc Univ, Brookhaven National Laboratory, pp 85–98, 1952.
13. Baker N, Shipley RA, Clark RE, Incefy GE, Skinner SS. $C^{14}$ studies in carbohydrate metabolism. V. Glucose metabolism in alloxan-diabetic rats. Amer J Physiol **200**:863–870, 1961.
14. Depocas F, Masironi R. Body glucose as fuel for thermogenesis in the white rat exposed to cold. Amer J Physiol **199**:1051–1055, 1960.
15. Feller DD, Neville ED. Insulin-like effect of bovine growth hormone in vivo as demonstrated by oxidation of $^{14}C$-U-glucose in diabetic rats. Physiol Chem Phys **10**:291–304, 1978.
16. Feller DD, Strisower EH, Chaikoff IL. Turnover and oxidation of body glucose in normal and alloxan-diabetic rats. J Biol Chem **187**:571–588, 1950.
17. Steenrod WJ Jr, Prahl JW, Barron EJ. Metabolism of lactate in alloxan diabetic acidosis. Diabetes **15**:423–429, 1966.
18. Drury DR, Wick AN. Metabolism of lactic acid in the intact rabbit. Amer J Physiol **184**:304–308, 1956.
19. Forbath N, Kenshole AB, Hetenyi G Jr. Turnover of lactic acid in normal and diabetic dogs calculated by two tracer methods. Amer J Physiol **212**:1179–1184, 1967.
20. Searle GL, Strisower EH, Chaikoff EL. Determination of rates of glucose oxidation in normal and diabetic dogs by a technique involving continuous injection of $C^{14}$-glucose. Amer J Physiol **185**:589–594, 1956.
21. Steele R, Altszuler N, Wall JS, Dunn A, deBodo C. Influence of adrenalectomy on glucose turnover and conversion to $CO_2$: studies with $C^{14}$ glucose in the dog. Amer J Physiol **196**:221–230, 1959.
22. Annison EF, Linzell JL. The oxidation and utilization of glucose and acetate by the mammary gland of the goat in relation to their overall metabolism and to milk formation. J Physiol **175**:372–385, 1964.
23. Annison EF, Lindsay DB, White RR. Metabolic interrelations of glucose and lactate in sheep. Biochem J **88**:243–248, 1963.
24. Bergman EN. Quantitative aspects of glucose metabolism in pregnant and nonpregnant sheep. Amer J Physiol **204**:147–152, 1963.
25. Bergman EN, Brockman RP, Kaufman CF. Glucose metabolism in ruminants: comparison of whole-body turnover with production by gut, liver and kidneys. Fed Proc **33**:1849–1454, 1974.
26. Searle GL, and Cavalieri RR. Determination of lactate kinetics in the human analysis of data from single injection vs continuous infusion methods. Proc Soc Exp Biol Med **139**:1002–1006, 1971.
27. Mukherjee SP, Mukherjee C, Yeager MD, Lynn WS. Stimulation of glucose transport and oxidation in adipocytes by fatty acids: evidence for a regulatory role in the cellular response to insulin. Biochem Biophys Res Commun **94**:682–289, 1980.
28. Rodbell M. Metabolism of isolated fat cells. I. Effects of hormones on glucose metabolism and lipolysis. J Biol Chem **239**:375–380, 1964.
29. Philipps AF, Carson BS, Meschia G, Battaglia FC. Insulin secretion in fetal and newborn sheep. Amer J Physiol **235**:E34–E38, 1978.
30. Boyd, RDH, Morriss FH Jr, Meschia G, Makowski EL, Battaglia FC. Growth of glucose and oxygen uptake by fetuses of fed and starved ewes. *Amer J Physiol* **225**:897, 1973.

31. James EJ, Meschia G, Battaglia FC. A-V differences of free fatty acids and glycerol in the ovine umbilical circulation. Proc Soc Exp Biol **138**:823–826, 1971.
32. Widdowson EM. Chemical Composition of newly born mammals. Nature (London) **166**:626–628, 1950.
33. Char VC, Creasy RK. Acetate as a metabolic substrate in the fetal lamb. Amer J Physiol **230**:357–361, 1976.
34. Morriss FH Jr, Boyd RDH, Makowski EL, Meschia G, Battaglia FC. Umbilical V-A differences of acetoacetate and betahydroxybutyrate in fed and starved ewes. Proc Soc Exp Biol Med **145**:879–883, 1974.
35. Meier PR, Teng C, Battaglia FC, Meschia G. The rate of amino acid nitrogen and total nitrogen accumulation in the fetal lamb. Proc Soc Exp Biol Med **167**:463–468, 1981.
36. Lemons JA, Adcock EW III, Jones MD Jr, Naughton MA, Meschia G, Battaglia FC. Umbilical uptake of amino acids in the unstressed fetal lamb. J Clin Invest **58**:1428–1434, 1976.
37. Meier PR, Peterson RG, Bonds DR, Meschia G, Battaglia FC. Rates of protein synthesis and turnover in fetal life. Amer J Physiol **240**:E320–E324, 1981.

---

Received January 11, 1983. P.S.E.B.M. 1983, Vol. 173.